Standard Operating Procedure for Particulate-Phase Total Nitrogen by Alkaline Persulfate Oxidation Digestion (Lachat Method)

LG208

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1.0 SCOPE AND APPLICATION

1.1 This method covers the determination of the various species of nitrogen compounds, excluding nitrogen gas, in particulate, matrices oxidized to nitrate by alkaline potassium persulfate digestion.

1.2 The approximate working range is 0.03 to 2.00 mg N/L.

2.0 SUMMARY

2.1 Nitrogen compounds are oxidized to nitrogen in alkaline persulfate spontaneously when the media is autoclaved under 15 psi and 121°C. Nitrogen losses are not observed when oxidation occurs under these pressure conditions.

2.2 Nitrate is quantitatively reduced to nitrite by passage of the sample through a column containing copper coated cadmium. The nitrite (reduced nitrate plus original nitrite) is determined by diazotizing with sulfanilamide dihydrochloride. The resulting water soluble dye has a magenta color which is read at 520 nm.

3.0 SAMPLE HANDLING AND COLLECTION

3.1 Samples are filtered by vacuum through 47 mm cellulose acetate filters with a 0.45 µm pore size. The filters retaining particles are collected into clean Petri dishes and frozen until analysis.

4.0 INTERFERENCES

4.1 Residual chlorine can interfere by oxidizing the cadmium column.

4.2 A major interference can come from ammonia contamination of glassware or reagents. To prevent this, glassware and utensils should be washed in 1 N hydrochloric acid and stored in an ammonia-free environment. Use only high purity potassium persulfate and store in a dry ammonia-free environment.

4.3 The total carbon content of the sample in excess of 20 mg C/L may interfere with the completion of the alkaline persulfate oxidation.

5.0 APPARATUS

5.1 60-mL test tubes with polypropylene Teflon-lined screw caps

5.2 Lachat QuikChem AE

5.2.1 XYZ Sampler

5.2.2 Nitrate/Nitrite Manifold (Lachat Method # 31-107-04-1-B)

5.2.3 Cadmium-Copper Reduction Column

5.2.4 Printer

NOTE: *Replacement cadmium columns are obtained from Lachat Instruments.*

5.3 Autoclave

6.0 REAGENTS AND STANDARDS

6.1 All reagents should be stored in the appropriate bottles and labeled with the following information:

Identity:	(15 N Sodium hydroxide)
Date of Preparation:	(mm/dd/yy)
Expiration Date:	(mm/dd/yy)
Concentration:	(1000 mgN/L)
Initials of Preparer:	(M.S.)

6.2 **Use reagent water for all solutions.**

6.3 **15 N Sodium Hydroxide:** Add 150 g of NaOH to 250 mL of reagent water.

CAUTION: *The solution will get very hot!* Swirl until dissolved. Cool and store in a plastic bottle.

6.4 **Ammonium Hydroxide Buffer, pH 8.5:** In a fume hood, to a 1-L volumetric flask containing approximately 250 mL of reagent water, add 105 mL concentrated hydrochloric acid (HCl). Carefully add 95 mL of ammonium hydroxide (NH_4OH). Stir very carefully. This liquid fumes and may spew. Add 1.0 g disodium ethylene diamine tetra acetic acid dihydrate ($Na_2EDTA•2H_20$). Adjust pH to 8.5 with 15 N NaOH and dilute to 1 L. Degas with Helium prior to each use.

6.5 **Sulfanilamide Color Reagent:** To a 1-L volumetric flask add approximately 600 mL of reagent water, 100 mL of 85% phosphoric acid, 40.0 g sulfanilamide, and 1.0 g N (1-naphthyl) ethylene diamine dihydrochloride (NED). Shake to wet, and stir to dissolve. Dilute to the mark, and invert to mix. Store in a dark bottle. This solution is stable for one month. Degas with Helium prior to each use.

6.6 **Alkaline Persulfate Digestion Solution:** In a 200-mL volumetric flask, dissolve 10.0 g potassium persulfate ($K_2S_2O_8$), and 1.5 g sodium hydroxide in approximately 150 mL reagent water and dilute to the mark. Prepare fresh weekly. Store on plastic container at room temperature.

6.7 **Borate Buffer, 1.0 M, pH 7.5:** In a 200-mL volumetric flask, dissolve 12.4 g boric acid, 1.6 g sodium hydroxide in about 150 mL of reagent water. Mix the solution on a magnetic stirrer and dilute to the mark.

6.8 Preparation of Calibration Standards

6.8.1 **Stock 1000 mgN/L Nitrate Solution:** Dissolve 7.218 g of potassium nitrate (KNO_3), dried for 1 hour at 105°C, in 500 mL of reagent water and dilute to 1 L.

6.8.2 **Intermediate 100 mgN/L Nitrate Standard Solution:** Dilute 100 mL of Stock Nitrate Solution (6.8.1) to 1 L with reagent water.

6.8.3 **Working Standards:** Prepare standards over the range of analysis. For the working range of 0 - 2.00 mg N/L, the following standards may be used:

mL of Intermediate Standard Solution (6.8.2) diluted to 1 L	Concentration in mg N/L
0.0	0.0
1.0	0.10
2.5	0.25
5.0	0.50
7.5	0.75
10.0	1.00
20.0	2.00

6.9 Preparation of Control Standards

6.9.1 **Stock 100 mg N/L Nitrate Control Standards:** Any nitrate compound may be used for control standards. The control standards should be prepared by someone other than the analyst. Weigh 0.5359 g of glycine ($NH_2CH_2CO_2H$), dried at 75°C for 1 hour, and dissolve in 500 mL of reagent water. Dilute to 1 L in volumetric flask with reagent water. The **Organic Spike** is prepared by adding 0.200 mL of the 6.9.1 standard to 36 mL of sample.

6.9.2 Prepare the control standards using solution (6.9.1).

QC Type	mL Control Standard Solution (6.9.1) diluted to 1 Liter	Concentration in mg N/L
Low Check Standard (CL)	4	0.40
High Check Standard (CH)	12	1.20

7.0 PROCEDURE

7.1 Digestion Procedure

7.1.1 The filters should be carefully transferred to the digestion tubes. Add 36 mL of reagent water and 4 mL of the digestion solution (6.6) to the tube. Recap the tube.

7.1.2 To cleansed, dried tubes, add 36 mL of working calibration standards, control standards, and blanks. Add 4 mL of digestion solution (6.6) to each tube and close with cap.

7.1.3 Autoclave samples and standards for 45 minutes at 15 psi at 121°C. Bring samples and standards to the room temperature and add 1 mL of borate buffer (6.7).

7.2 Follow the Lachat Procedural SOP.

8.0 CALCULATIONS

8.1 The computer yields results in mg N (as NO_2+NO_3)/L.

8.2 Particulate Total Nitrogen Result Calculation

$$C_{PTN} = \frac{C_N}{25 \times V}$$

Where:

C_{PTN} = Concentration of particulate total nitrogen in water (mg/L)
C_N = Concentration of nitrogen from LACHAT instrument (mg/L)
V = Volume of filtered water (L)

9.0 QUALITY CONTROL

9.1 Refer to the Chapter 2 Introduction for definitions of quality control samples and information regarding quality control procedures, such as QC sample IDs and labeling.

9.2 The following QC samples must be prepared and analyzed at the minimum frequency indicated.

QC Sample Type		Frequency	Acceptance Criteria
External	Field Reagent Blank (FRB)	One per basin[a]	0.00 ± 0.04 mg/L or less than one tenth associated field sample concentrations, whichever is greater
	Lab Duplicate (LD1)	One per basin[a]	RPD ≤ 20%
Internal	Calibration	At the beginning of each batch	r ≤ 0.995
	High Check Standard (CH)	At the beginning & end of each batch or 1 per 40 samples, whichever is more frequent	1.20 ± 0.12 mg/L
	Low Check Standard (CL)	At the beginning & end of each batch or 1 per 40 samples, whichever is more frequent	0.40 ± 0.04 mg/L
	Laboratory Reagent Blank (LRB)	At the beginning & end of each batch or 1 per 40 samples, whichever is more frequent	0.00 ± 0.04 mg/L
	Matrix Spike (MS)	During each batch or 1 per 40 samples, whichever is more frequent	100% ± 20%
	Method Detection Limit (MDL)	Once per year and each time a significant change is made to the SOP	0.03 mg/L

[a] A field duplicate, lab duplicate, and field reagent blank are collected with each group of 3, 4, or 5 stations depending on the lake. A Random Number Generator (RNG) is used to determine the stations and depths of these QC samples. Where basins are well defined, at least one of each is collected from each basin.

9.3 Assessment

9.3.1 The analyst must compare analytical results to the acceptance criteria listed in Section 9.2 to identify QC failures. If the results are outside the acceptance criteria, the analyst should first review their calculations for errors and if none are identified, they must follow the corrective action procedures listed in Section 9.4.

9.4 Corrective Actions

9.4.1 Corrective action procedures will often be handled at the bench level by the analyst, who reviews the preparation or extraction procedure for possible errors, checks the instrument calibration, spike and calibration mixes, instrument sensitivity, and any other potential sources of error. If failure occurs and an error is identified, the analyst should re-run quality control and RFS samples in the entire analytical batch to confirm the results. Because external QC samples are collected and prepared during the survey and provided to the contractor or grantee laboratory, a single rerun to confirm results is sufficient when all other QC samples are within acceptance criteria. For analyses conducted onboard, if the problem persists or cannot be identified, the matter must be referred to the Chief Scientist for further investigation. Depending upon the Chief Scientist's evaluation, the analyst may or may not be required to prepare and re-run the samples again. Additionally, if the results are significantly different than the expected concentrations based on historical data or related samples, then the analyst may split the RFS sample in the laboratory and analyze the splits. Once a decision is made, full documentation of the corrective action procedures and assessment of the final result must be filed with the WQS QM Technical Lead (Marvin Palmer) or the GLNPO QM. For analyses conducted at contract or grantee laboratories, this information can be included with submitted data. When contractor or grantee laboratories have a question regarding acceptable corrective actions, they should contact the Biology Technical Lead or Limnology Technical Lead as appropriate for instruction at a time when corrective action can still be taken.

9.5 Data Reporting/Recording

9.5.1 When corrective actions are not feasible or do not resolve QC failure, the analyst is responsible for identifying all failed QC samples and RFS samples. If analyses are being conducted onboard, the analyst should document the QC information on the hard-copy Field Information Recording Forms (Appendix H. If analyses are being conducted by contract or grantee laboratories, the analyst should document all QC information with the submitted data.

10.0 WASTE DISPOSAL

10.1 Effluent from this analysis should be neutralized with sodium hydroxide to a pH of 6 - 9 and then washed down the laboratory drain with plenty of water.

11.0 PREVENTATIVE MAINTENANCE

11.1 Required maintenance is described in the Lachat Procedural SOP.

12.0 TROUBLESHOOTING

12.1 The most common problem is deactivation of the cadmium column, which result in, low values and non-linear calibration curves. The deactivation of the column is quantified by a column having less than a 90% efficiency factor. To determine cadmium column efficiency, prepare a 0.600 mg N as NO_2 /L solution.

12.2 Preparation of the Sodium Nitrite Column Efficiency Test Standards

12.2.1 To prepare the 100 mg N (as NO_2^-) Nitrite Stock Standard, dissolve 0.4926 g Sodium Nitrite ($NaNO_2$) dried for 1 hour at 105°C. in 500 mL of reagent water. Add 1 mL of H_2SO_4 (conc.) and dilute to 1 L. Mix this solution thoroughly.

12.2.2 From the 12.2.1 Stock solution, take 6.0 mL and add this to 500 mL of reagent water. Add 1 mL of H_2SO_4 (conc.) and dilute to 1 L. Concentration of this solution is 0.60 mg N (as NO_2^-)/L. Store in a plastic container at 4°C.

12.2.3 From the 6.9.2 Stock solution, take 6.0 mL, and add this to 500 mL of reagent water. Add 1 mL of H_2SO_4 (conc.) and dilute to 1 L. Concentration of this solution is 0.60 N (as NO_3^-)/L. Store in a plastic container at 4°C.

12.3 Column efficiency test

12.3.1 Calibrate instrument with calibration standards.

12.3.2 Analyze NO_2^- standard (12.2.2).

12.3.3 Analyze the matching concentration NO_3^- standard (12.2.3).

12.3.4 Cadmium column efficiency is calculated by comparing the known concentration of the 0.60 mg N (as NO_3^-)/L (12.2.3) to the known concentration of 0.60 mg N (as NO_2^-)/L (12.2.2). The acceptable efficiency is ≥ 90%.

$$Percent\ Efficiency = \frac{Conc.\ NO_3^-}{Conc.\ NO_2^-} \times 100\%$$

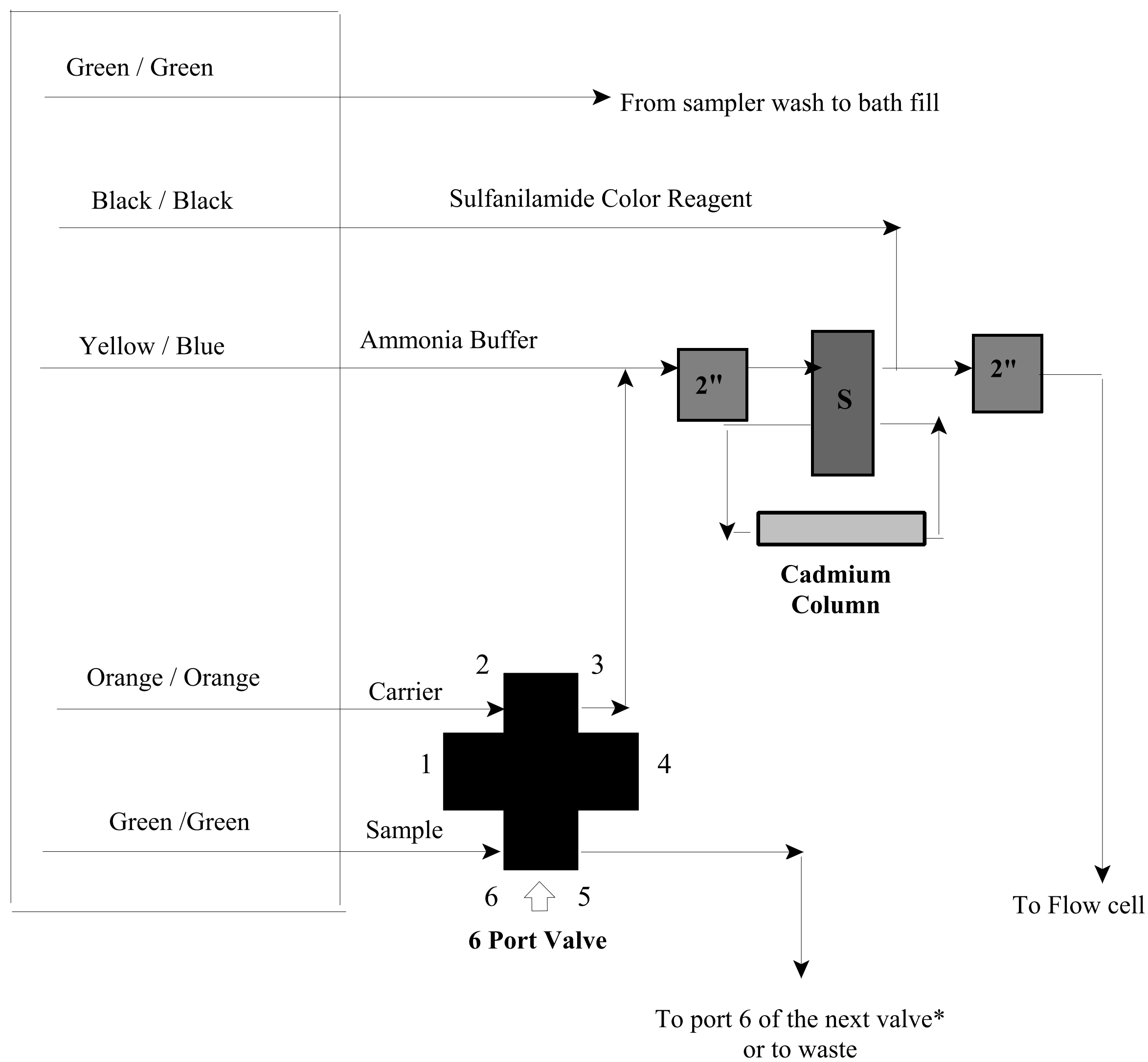
12.4 The efficiency test should be performed monthly.

13.0 REFERENCES

13.1 Lachat Instruments, Method Number 10-107-04-1-C, Nitrate/Nitrite in Surface Water, Wastewater, Revision Date November 1992.

13.2 Lachat QuikChem AE Operating Manual.

14.0 NITRATE - NITRITE ANALYTICAL MANIFOLD



Legend

2" - 4" Mixing coil (there is 135 cm of tubing on the 4.0" coil support)

S - This is a 2 state switching valve used to place the cadmium column in-lane with the manifold

Comments
1. Filter used is 520 nm.
2. Sample loop length is 22.5 cm. (0.032") ID.
3. All manifold tubing is 0.8 nm (0.032") ID. This related to a flow of 5.2 µL/cm.
4. The **carrier** is helium degased reagent water.

* If more than one channel is being used.